

Chapter 3. Microbiology

INTRODUCTION

The City of San Diego performs water quality monitoring along the shoreline and in offshore ocean waters for the region surrounding the Point Loma Ocean Outfall (PLOO). This aspect of the City's ocean monitoring program is designed to assess general oceanographic conditions, evaluate patterns in movement and dispersal of the PLOO wastewater plume, and monitor compliance with water contact standards defined in the 2001 California Ocean Plan (COP) as according to NPDES permit specifications (see Chapter 1). Results of all sampling and analyses, including COP compliance summaries, are submitted to the San Diego Regional Water Quality Control Board in the form of monthly receiving waters monitoring reports. Densities of indicator bacteria (total coliforms, fecal coliforms, enterococcus), along with oceanographic data (see Chapter 2), are evaluated to provide information about the movement and dispersion of wastewater discharged to the Pacific Ocean through the outfall. Analyses of these data may also help identify other point or non-point sources of bacterial contamination in the region (e.g., outflows from rivers or bays, surface runoff from local watersheds). This chapter summarizes and interprets patterns in seawater bacterial concentrations collected for the Point Loma region during 2007.

MATERIALS AND METHODS

Field Sampling

Seawater samples for bacteriological analyses were collected at a total of 52 NPDES-mandated shore, kelp bed or offshore monitoring sites during 2007 (**Figure 3.1**). Sampling was performed weekly at eight monitoring sites located along the shore (i.e., stations D4, D5, and D7–D12) to monitor bacterial levels along public beaches and evaluate compliance with the 2001 COP water contact standards (see **Box 3.1**). Eight stations located

in nearshore waters within the Point Loma kelp forest were also monitored to assess water quality conditions in areas used for recreational activities such as SCUBA diving, surfing, fishing and kayaking. These include stations C4, C5 and C6 located near the inner edge of the kelp bed along the 9-m depth contour, and stations A1, A6, A7, C7 and C8 located near the outer edge of the kelp bed along the 18-m depth contour. The kelp stations are also subject to COP water contact standards, and were therefore sampled weekly, such that each day of the week was represented over a two month period. Thirty-six offshore stations (F01–F36) were sampled quarterly during January, April, July and October in order to estimate the spatial extent of the wastewater plume at these times. Complete sampling of all 36 stations usually occurs over a 3 day period. Three of these offshore sites (stations F01–F03) are located along the 18-m depth contour, while 33 sites

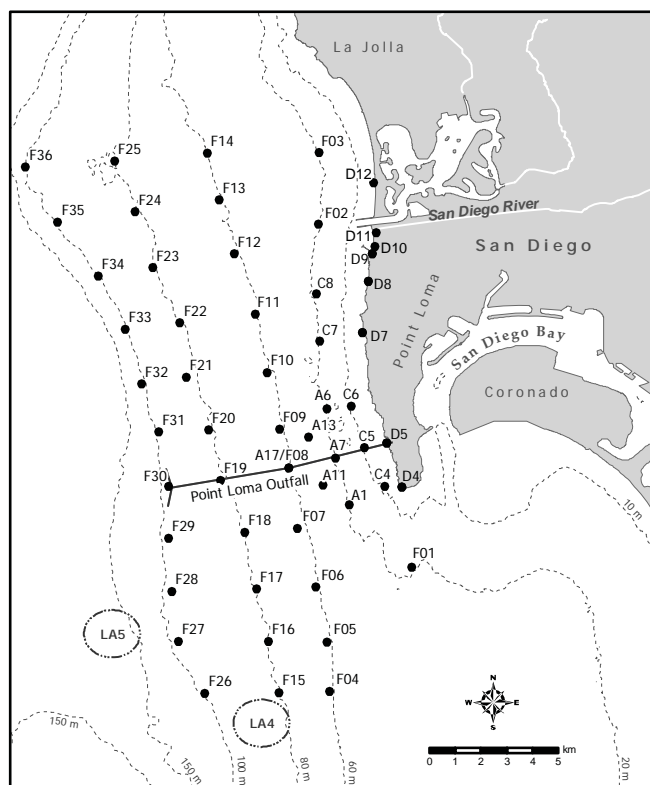


Figure 3.1

Water quality monitoring stations for the Point Loma Ocean Outfall Monitoring Program.

Box 3.1

Bacteriological compliance standards for water contact areas, 2001 California Ocean Plan (SWRCB 2001). CFU = colony forming units.

- (1) *30-day total coliform standard* — no more than 20% of the samples at a given station in any 30-day period may exceed a concentration of 1000 CFU per 100 mL.
- (2) *10,000 total coliform standard* — no single sample, when verified by a repeat sample collected within 48 hrs, may exceed a concentration of 10,000 CFU per 100 mL.
- (3) *60-day fecal coliform standard* — no more than 10% of the samples at a given station in any 60-day period may exceed a concentration of 400 CFU per 100 mL.
- (4) *geometric mean* — the geometric mean of the fecal coliform concentration at any given station in any 30-day period may not exceed 200 CFU per 100 mL, based on no fewer than 5 samples.

(11 per transect) are distributed along the 60-m (stations F04–F14), 80-m (stations F15–F25) and 98-m (stations F26–F36) depth contours. In addition, three sites (stations A11, A13, A17) located seaward of the kelp bed were sampled as part of the weekly kelp bed sampling array to ensure that water quality is appropriately documented in the area of the original Point Loma discharge location. Analyses for these additional special study stations are not included herein, but have been reported previously (see City of San Diego 2007b, 2008a).

Seawater samples for the eight shore stations were collected from the surf zone and stored in sterile 250-mL bottles. In addition, visual observations of water color and clarity, surf height, human or animal activity, and weather conditions were recorded at the time of sample collection. The samples were then transported on blue ice to the City of San Diego's Marine Microbiology Laboratory (CSDMML) and analyzed to determine concentrations of total coliform, fecal coliform, and enterococcus bacteria. Seawater samples from the kelp bed and quarterly offshore stations were collected using either a series of Van Dorn bottles or a rosette sampler fitted with Niskin bottles. These samples were collected at 3-5 discrete depths per site dependent upon station depth (see **Table 3.1**). Aliquots for each analysis were drawn into appropriate sample containers. All seawater samples were refrigerated on board

ship and then transported to the CSDMML for analysis of the above indicator bacteria (total and fecal coliforms, enterococcus). Visual observations of weather conditions, sea state, and human or animal activity in the area were also recorded at the time of sampling. Monitoring of the PLOO area and neighboring coastline also included aerial and satellite image analysis performed by Ocean Imaging of Solana Beach, California (Svejkovsky 2008; also see Chapter 2).

Laboratory Analyses and Data Treatment

All bacterial analyses were performed within 8 hours of sample collection and conformed to the standard membrane filtration techniques (see APHA 1992). The CSDMML follows guidelines issued by the EPA Water Quality Office, Water Hygiene Division, and the California State Department of Health Services (CDHS) Environmental Laboratory Accreditation Program (ELAP) with respect to sampling and analytical procedures (Bordner et al. 1978, APHA 1992).

Colony counting of indicator bacteria, calculation of results, data verification and reporting all follow guidelines established by the EPA (Bordner et al. 1978). According to these guidelines, plates with bacterial counts above or below the ideal counting range were given greater than (>), less

Table 3.1

Depths at which bacteriological samples are collected at the PLOO kelp and quarterly offshore stations.

Station transect	Sample depth (m)								
	1	3	9	12	18	25	60	80	98
9-m Kelp bed	x	x	x						
18-m Kelp bed	x			x	x				
18-m Offshore	x			x	x				
60-m Offshore	x					x	x		
80-m Offshore	x					x	x	x	
98-m Offshore	x					x	x	x	x

than (<), or estimated (e) qualifiers. However, these qualifiers were dropped and the counts treated as discrete values during calculation of mean values and in determining compliance with COP standards.

Bacteriological benchmarks defined in the 2001 COP or Assembly Bill 411 (AB 411) were used as reference points to distinguish elevated bacteriological values in receiving water samples discussed in this report. These benchmarks are: (a) >1000 CFU/100 mL for total coliforms; (b) >400 CFU/100 mL for fecal coliforms; (c) >104 CFU/100 mL for enterococcus. Furthermore, any seawater sample with a total coliform concentration ≥ 1000 CFU/100 mL and a fecal:total (F:T) ratio ≥ 0.1 is considered representative of contaminated waters (see CDHS 2000). Samples that met these latter two criteria were used as indicators of the PLOO waste field or other sources of bacterial contamination.

Quality assurance tests were performed routinely on seawater samples to ensure that sampling variability did not exceed acceptable limits. Duplicate and split bacteriological samples were collected and processed according to method requirements to measure intra-sample and inter-analyst variability, respectively. Results of these procedures were reported in the laboratory's Quality Assurance Report for 2007 (City of San Diego 2008b).

Maps of total coliform densities at the offshore stations were generated using the Spatial Analyst extension for ArcGIS 9.1 in order to estimate possible distribution of the PLOO waste field during the quarterly sampling months. The Inverse Distance

Weighting algorithm was used with the power set to 3, a neighborhood of 5, and default values for all other parameters. Coliform densities from samples collected at depths shallower than 60 m were not used because contaminated water was detected in only four such samples during the year. The interpolations of coliform distribution patterns in these relatively deep waters are meant for simplified data visualization purposes only and are not statistically significant.

RESULTS AND DISCUSSION

Shore Stations

Concentrations of indicator bacteria were generally very low along the shoreline in 2007, which likely reflects the relatively low rainfall that occurred during the year (see **Table 3.2**). Monthly densities at the different shore stations averaged 24–622 CFU/100 mL for total coliforms, 4–224 CFU/100 mL for fecal coliforms, and 4–215 CFU/100 mL for enterococcus. Five of the nine shoreline samples that had total coliform concentrations $\geq 1,000$ CFU/100 mL during the year were collected within 72 hours after a rain event (see **Table 3.3**). These included samples collected at stations D9, D10 or D11 during February, April or December. Three of the five samples also had F:T ratios ≥ 0.1 and were therefore indicative of contaminated waters (i.e., samples from D11 in February, and from D10 and D11 in December). In contrast, the other four samples with elevated total coliforms occurred during periods of no rain. These included samples collected at stations D4 and D11 in June, station D7 in July, and station D8

Table 3.2

Summary of rainfall and indicator bacteria levels at PLOO shore stations during 2007. Rainfall data are from Lindbergh Field, San Diego, CA. Total coliform (Total), fecal coliform (Fecal), and enterococcus (Entero) densities are expressed as mean CFU/100 mL per month. Stations are listed from south to north.

Month	Rain (in)		D4	D5	D7	D8	D9	D10	D11	D12	All stations
Jan	0.51	<i>Total</i>	29	21	14	204	4	18	21	19	41
		<i>Fecal</i>	3	3	2	26	3	14	10	8	9
		<i>Entero</i>	5	14	4	26	2	5	6	20	10
Feb	1.12	<i>Total</i>	5	3	3	102	27	224	393	22	98
		<i>Fecal</i>	2	2	2	8	4	58	50	11	17
		<i>Entero</i>	3	2	2	29	5	52	38	17	18
Mar	0.09	<i>Total</i>	2	5	5	101	5	18	50	8	24
		<i>Fecal</i>	2	2	3	7	2	3	9	3	4
		<i>Entero</i>	2	2	2	7	2	21	5	4	6
Apr	0.46	<i>Total</i>	6	13	4	28	253	51	222	15	74
		<i>Fecal</i>	4	2	3	9	3	14	24	6	8
		<i>Entero</i>	2	2	3	8	4	7	10	137	22
May	0.00	<i>Total</i>	10	11	49	21	16	64	36	2	26
		<i>Fecal</i>	4	2	2	3	2	6	21	6	6
		<i>Entero</i>	2	2	2	3	4	6	9	4	4
Jun	0.00	<i>Total</i>	1085	18	132	76	20	100	256	56	218
		<i>Fecal</i>	42	6	2	34	2	62	207	4	45
		<i>Entero</i>	2	12	3	2	2	12	1686	2	215
Jul	0.00	<i>Total</i>	20	56	696	20	64	88	78	16	130
		<i>Fecal</i>	7	2	283	3	6	30	13	2	43
		<i>Entero</i>	2	2	3	2	4	10	25	3	7
Aug	0.00	<i>Total</i>	132	92	128	132	132	64	36	90	101
		<i>Fecal</i>	3	2	5	14	3	13	22	2	8
		<i>Entero</i>	2	2	3	4	4	8	5	4	4
Sep	0.05	<i>Total</i>	50	70	47	47	25	38	36	31	43
		<i>Fecal</i>	3	2	5	7	3	16	9	10	7
		<i>Entero</i>	2	3	10	3	2	10	18	44	12
Oct	0.37	<i>Total</i>	16	92	128	604	49	58	26	16	124
		<i>Fecal</i>	3	8	10	185	2	12	4	4	29
		<i>Entero</i>	10	7	7	99	3	6	6	7	18
Nov	0.97	<i>Total</i>	13	28	52	164	17	21	13	40	43
		<i>Fecal</i>	3	11	9	64	4	11	7	20	16
		<i>Entero</i>	3	7	12	25	4	11	9	14	11
Dec	0.80	<i>Total</i>	39	25	20	132	71	2311	2331	45	622
		<i>Fecal</i>	2	2	10	38	11	811	894	20	224
		<i>Entero</i>	5	5	4	30	19	520	493	35	139
Annual means		n	64	64	62	62	62	62	62	61	
		Total	24	24	55	128	25	112	251	34	
		Fecal	3	5	12	48	5	21	24	8	
		Entero	3	6	6	46	5	13	27	11	

Table 3.3

Summary of samples with elevated total coliform (Total), fecal coliform (Fecal), and enterococcus (Entero) densities (CFU/100 mL) at PLOO shore stations in 2007. F:T = fecal total coliform ratio. Rainfall was measured at Lindbergh Field, San Diego, CA.

Date	72-Hour rain (in.)	Station	Total	Fecal	Entero	F:T
February 22	0.14	D11	1800	200	120	0.11
April 24	0.08	D11	1000	60	4	0.06
April 24	0.08	D9	1200	2	2	0.00
June 17	0.00	D4	5400	200	2	0.04
June 23	0.00	D11	1100	960	8400	0.87
July 29	0.00	D7	3200	1400	8	0.44
October 8	0.00	D8	1600	2	2	0.00
December 1	0.94	D10	>16,000	5600	3600	0.35
December 1	0.94	D11	>16,000	6200	3400	0.39

in October; the samples from D7 and D11 also had F:T values ≥ 0.1 . MODIS satellite imaging of the region on June 16, 17, 18, and 23 showed turbidity plumes from the San Diego River and San Diego Bay encompassing several of the shore stations (see Svejksky 2008), which may account for the elevated bacteria concentrations at stations D4 and D11. Furthermore, station D11 is located near the mouth of the San Diego River at a designated dog recreational area (Dog Beach). Contamination from both of these sources is a likely cause of the elevated bacterial counts at this shore station. A possible source of contamination at station D8 is a tidally influenced storm drain in the area (see Martin and Gruber 2005; City of San Diego 2007). Other sources that may have contributed to bacterial contamination along the shore include beach wrack (e.g., kelp and seagrass) and shorebirds, all of which were present during the collection of many seawater samples (City of San Diego 2007b, 2008a).

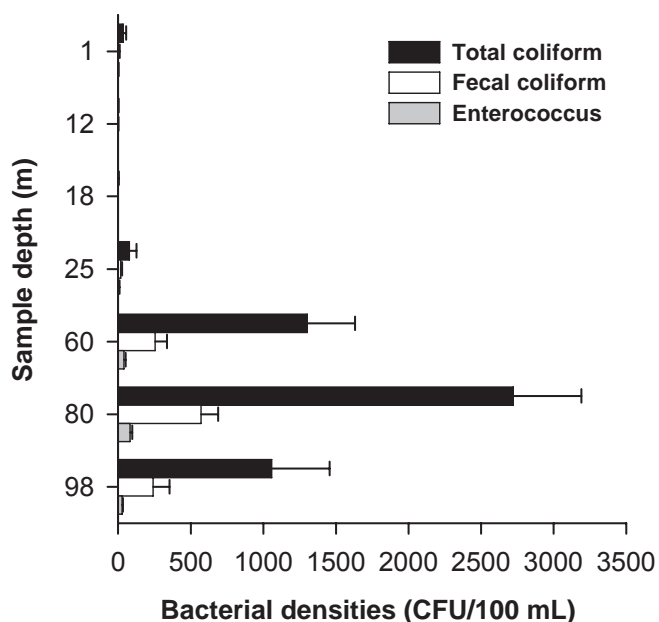
Kelp Stations

Concentrations of indicator bacteria were also very low at PLOO kelp stations in 2007. Densities at these sites during the year ranged from <2 to 3600 CFU/100 mL for total coliforms, <2 to 520 CFU/100 mL for fecal coliforms, and <2 to 110 CFU/100 mL for enterococcus (City of San Diego 2007b, 2008a). Of the 1,440 seawater samples taken from kelp stations, only four samples ($<1\%$) had elevated total coliform levels >1000 CFU/100 mL, of which only two were indicative of contaminated

seawater with F:T ratios ≥ 0.1 (Table 3.4). All four of the above samples were collected during a 72-hour rain event. No samples collected at the kelp stations had elevated fecal coliform or enterococcus values during the year.

Offshore Stations

A summary of bacterial densities collected at the PLOO offshore stations during 2007 is presented in Figure 3.2. Seawater samples collected from

**Figure 3.2**

Summary of bacterial densities at PLOO offshore stations sampled in 2007. Values are expressed as means \pm one standard deviation.

Table 3.4

Summary of samples with elevated total coliform (Total), fecal coliform (Fecal), and enterococcus (Entero) densities (CFU/100 mL) at PLOO kelp stations in 2007. F:T = fecal to total coliform ratio. Rainfall was measured at Lindbergh Field, San Diego, CA.

Date	72-Hour rain (in.)	Station	Depth (m)	Total	Fecal	Entero	F:T
March 1	0.15	A1	12	1400	280	34	0.20
March 1	0.15	A1	18	3600	400	78	0.11
March 1	0.15	A6	18	3600	280	88	0.08
March 1	0.15	A7	12	2000	44	100	0.02

shallow depths (i.e., along the 18-m depth contour) at the offshore stations had total coliform, fecal coliform, and enterococcus concentrations averaging less than 6 CFU/100 mL during 2007 (**Table 3.5**). In contrast, average densities of indicator bacteria from deeper waters were as high as 1831 CFU/100 mL for total coliforms (i.e., at 80 m during April). The highest average fecal coliforms (413 CFU/100 mL) also occurred in samples collected at a depth of 80 m, but during the month of January, while the highest mean enterococcus values (56 CFU/100 mL) occurred at depths of 80 and 98 m during January. Of the 564 samples collected, only 64 (~11%) may be considered indicative of contaminated waters with elevated total coliforms and an F:T ratio ≥ 0.1 (see **Appendix A.1**). Only four of these samples were collected from depths ≤ 25 m, including one sample from station F16 (25 m) and two samples from station F17 (1 m, 25 m) in April following a rain

event, and a single sample from station F08 (1 m) in October. All of the remaining contaminated samples were collected from depths of 60 m and greater. Overall, these results suggest that the wastewater plume was generally restricted to relatively deep waters throughout the year (see Chapter 2).

Interpolations of total coliform data from depths ≥ 60 suggest that the spatial distribution of the waste field varied by quarter in 2007 (**Figure 3.3**). During January, for example, the waste field appeared to be fairly well dispersed with the highest bacterial counts occurring at station F20 located to the northeast of the discharge area. The waste field was still well dispersed in April, although the highest bacterial counts appeared to be concentrated immediately around the discharge site (i.e., at station F30). In contrast, the wastewater plume appears to have generally dispersed to the north by

Table 3.5

Summary of indicator bacteria densities (CFU/100 mL) at PLOO offshore stations in 2007. Data for each quarterly survey are expressed as means for all stations along each depth contour; n = total number of samples.

Assay	Contour	n	January	April	July	October
<i>Total</i>	18-m offshore	9	6	2	4	2
	60-m offshore	33	47	129	10	170
	80-m offshore	44	1622	1831	678	205
	98-m offshore	55	1445	1461	1151	863
<i>Fecal</i>	18-m offshore	9	2	2	2	2
	60-m offshore	33	6	22	3	21
	80-m offshore	44	413	195	149	41
	98-m offshore	55	339	309	235	221
<i>Entero</i>	18-m offshore	9	2	2	2	2
	60-m offshore	33	4	5	2	10
	80-m offshore	44	56	34	17	13
	98-m offshore	55	56	46	40	19

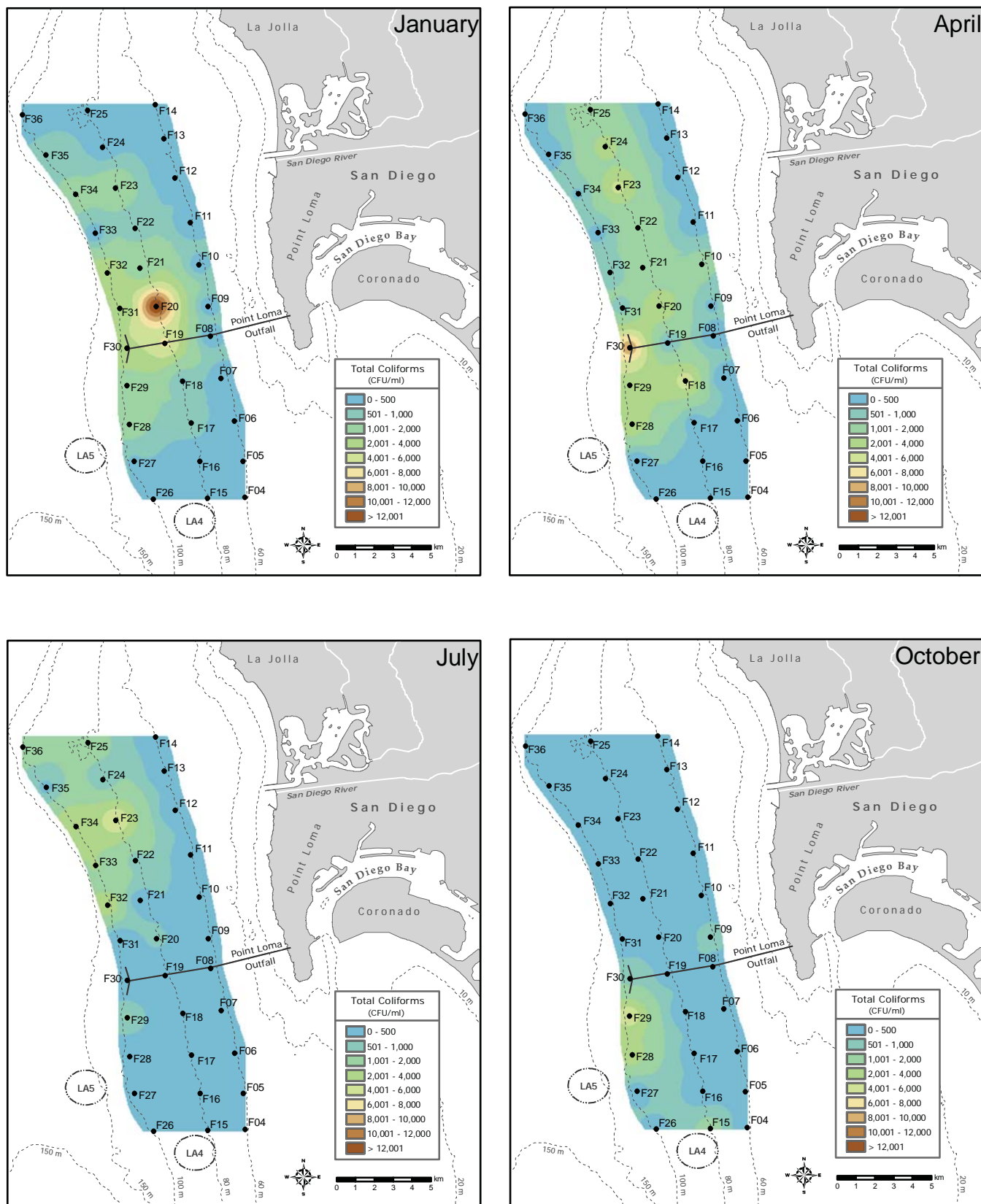


Figure 3.3

Total coliform distributions for seawater samples collected at depths ≥ 60 m during quarterly offshore surveys in 2007.

July. Finally, results for the October survey indicate that the plume reversed direction and had moved southward, which was consistent with MODIS imagery at the time which showed a southern flow at surface waters (see Svejksky 2008).

Compliance with California Ocean Plan Standards

Compliance with COP bacterial standards (Box 3.1) for the shore and kelp stations was very high in 2007 (**Appendices A.2, A.3**). Shore stations D8 and D11 were the only stations with seawater samples where bacteria levels fell below 100% compliance. Station D8, located near a tidally influenced storm drain, was 78% compliant with the 60-day fecal coliform standard, 92% compliant with the fecal geometric mean standard, and 100% compliant with the other two COP standards. Station D11, located near the mouth of the San Diego River, was 92% compliant with the 60-day fecal coliform standard and 100% compliant with the other three COP standards. All seawater samples collected from the kelp stations were 100% compliant with each COP bacterial standard.

SUMMARY AND CONCLUSIONS

There was no evidence that the Point Loma Ocean Outfall (PLOO) wastewater plume reached the shoreline or recreational waters in 2007. Elevated bacterial densities along the shore were limited to a few instances at stations D4, D7, D8, and D11 where the source of contamination may have been from rainfall, heavy recreational use, or decaying kelp and surfgrass wrack material. For example, most of elevated bacterial densities occurred during the wettest months of 2007 (i.e., February–May, December). Furthermore, all but two shore stations were 100% compliant with the four COP standards; these exceedances also corresponded with rainfall events or were associated with other sources of contamination unrelated to the PLOO.

It is unlikely that the PLOO wastewater ever reached surface waters in 2007. Bacteriological evidence

of contaminated water at the offshore stations was predominantly limited to samples collected from depths of 60 m and deeper. The discharge depth (~98 m) may be the dominant factor that keeps the plume from reaching the surface. Wastewater is released into cold, dense seawater that does not appear to mix with the top 25 m of the water column. Analysis of physical parameters during the year suggest that the water column was stratified during the spring through fall months (see Chapter 2). However, the absence of bacteriological contamination in surface waters during January, when the water column was well mixed, suggests that stratification may not be the only factor limiting the depth of the plume to 60 m and deeper.

The dominant direction of waste field flow appeared to be northward in 2007, except during October when it appeared to move in a southerly direction. High bacterial densities were detected at the northern limits of the quarterly sampling grid during most quarters, but only at the most southern sites in October.

Previous analyses of historical water quality data indicate that since the extension of the Point Loma outfall in 1993, the PLOO waste field no longer reaches the shoreline (City of San Diego 2007a). This pattern remained true for 2007 with evidence of the plume being restricted to mostly offshore waters at depths of 60 m or below.

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